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Perspective

A trait-based framework to identify microbial keystone taxa for microbiome engineering

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SUMMARY

Defining simplified microbial consortia and harnessing their functional potential holds promise for microbe-based solutions in agriculture, biotechnology, and medicine. However, defining their optimal composition remains challenging, primarily due to the vast taxonomic and functional diversity of natural microbiomes. Implementing the "keystone taxa" concept into microbiome research may help to define simplified consortia and prioritize microorganisms that drive essential ecosystem functions. The idea was developed in the 1960s to describe organisms with disproportionate ecological influence. Despite its potential, a systematic workflow for the identification of microbial keystone taxa has remained elusive. Here, we propose a trait-based 8-component framework that characterizes the ecological significance of microbial keystone taxa, and an operational 4-step approach to recover "keystone taxa candidates" from complex *omics* data and propose strategies for their *in vitro* and *in vivo* empirical assessments. This approach may facilitate the rational design of simplified microbial consortia with enhanced functional performance in both applied and natural contexts.

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INTRODUCTION

Nature provides ecosystem services that are essential for life on Earth. Direct interaction with nature significantly impacts human health, influencing well-being and disease prevalence.[1] However, human activities have dramatically changed our environment, resulting in habitat destruction, biodiversity loss, pollution, climate change, overexploitation of species, and the introduction of invasive organisms.[2] These changes have been considered potential causes for notable increases in environment-related diseases in recent years, including infections, cancer, allergies, and metabolic disorders.[3] The growing awareness of the connection between environmental and human health is reflected in the relatively recent development of holistic concepts, such as One Health, Planetary Health, or Global Health.[4] In these concepts, environmental microbes and their functional potential are recognized as architects of the ecosystem services provided by nature. Therefore, changes in the environmental microbiome or losses of diversity have been considered a reason for losses of ecosystem services and reduced resilience toward environmental stressors. Environmental microbes act as a reservoir from which humans recruit beneficial species through food, water, soil, and physical contact. For example, bacteria originally associated with plants and soil, such as *Lactobacillus* or *Bifidobacterium*, have been domesticated through food fermentation and now form key members of the human gut microbiome.[5] Thus, changes in the environmental microbiome can be mirrored in the human microbiome, potentially leading to dysbiosis, which refers to a state of microbial imbalance and functional disruption that has been linked to various diseases.[6] Consequently, preventing or mitigating the loss of microbial diversity in nature is crucial for promoting both environmental and human health.

In recent years, a growing field of research has focused on compensating for microbial losses through the targeted reintroduction of microbiota into affected ecosystems, either as single strains or simplified microbial consortia.[7] This approach has gained significant interest, for example, in agriculture, where "biostimulants" (i.e., environmental probiotics) have been proven to reduce the use of pesticides and inorganic fertilizers.[8] In human medicine, losses in microbial diversity, e.g., after antibiotic therapy, are compensated by using beneficial microbiota (i.e., probiotics) to reestablish a healthy gut microbiome.[9] Recently, industrial biotechnology has also started to implement simplified microbial consortia in fermentation and bioprocesses, which can improve bioproduction yields.[10]

The successful application of simplified microbial consortia is context-dependent, often limited by the fact that the needed microbes often remain uncultured due to their specific physiological requirements or the need for a minimal microbial consortium for their successful establishment.[10] Thus, a vast part of current microbiome research aims to develop targeted isolation approaches, using new *omics* techniques.[11] However, this can be very challenging, considering the vast microbial diversity in most ecosystems. For example, one gram of soil may harbor more than 54,000 different microbial species,[12] the human gut over 5,600 bacterial species,[13] and marine environments an estimated ~1.3 million prokaryotic species (www.marinespecies.org). The lack of information regarding their unique eco-physiological needs hinders the successful isolation of most microbial diversity based on current technologies. It is therefore essential to strategically prioritize the most relevant taxa for isolation and simplified microbial consortia development. Adapting the "keystone taxa" concept from classical ecology can guide this process by focusing on microbes with the greatest functional influence on their communities. An operational framework for the systematic identification of such influential taxa could facilitate targeted enrichment and integration into simplified microbial consortia, ultimately enhancing the efficiency, success, and stability of microbiome-based applications. Indeed, several studies have already shown that keystone members can be systematically identified and that their presence strongly influences community function, which suggests their potential for designing more effective simplified microbial consortia: one study showed that removal of a single strain from a maize root synthetic community caused collapse of community function[14]; another revealed keystone effects shaping assembly in the Arabidopsis phyllosphere[15]; and a more recent work reported that native keystone taxa enhanced pathogen suppression in tomato rhizospheres.[16]

Here, we present a trait-based approach that captures essential characteristics of microbial keystone taxa and their ecological interactions, facilitating the understanding of their ecological significance and eco-physiological requirements for isolation or enrichment. We present a flexible, multi-dimensional framework of diagnostic traits to aid in the identification and prioritization of microbial keystone taxa across diverse ecosystems (summarized in Figure 1 and Box 1). Additionally, we outline a 4-step operational approach (Figure 2) for detecting candidate keystone taxa in large-scale *omics* datasets and propose strategies for their targeted isolation. Although we focus here on taxa that promote beneficial functional patterns across ecosystems for biotechnological applications, the framework can also be applied to negative interactions such as dysbiosis, invasion, or disease complexes.

HOW DO WE DEFINE MICROBIAL KEYSTONE TAXA?

Since Paine introduced the idea of "keystone species" in 1969,[19] this concept has been a central point of interest in ecology and related fields. At that time, Paine was studying a shore ecosystem, and he realized that the presence of one particular species could mostly control the ecosystem structure and functioning. That species was the sea star, and he labeled this species a keystone species for that ecosystem.[19] In the following years, Paine and colleagues generalized this finding and proposed that keystone species disproportionately impact their community or ecosystem relative to their abundance[20] (Figure 1, high impact regardless of abundance). Since then, the term keystone species has been broadly used in the ecology of macroorganisms, especially in food-web studies.[21]

More recently, the keystone species concept was extended to microbiology in line with sequencing technology and computational infrastructure developments. The current definition of *microbial keystone taxa* refers to specific microorganisms within a microbial community that play critical roles in maintaining the overall structure, function, and stability of a given microbial

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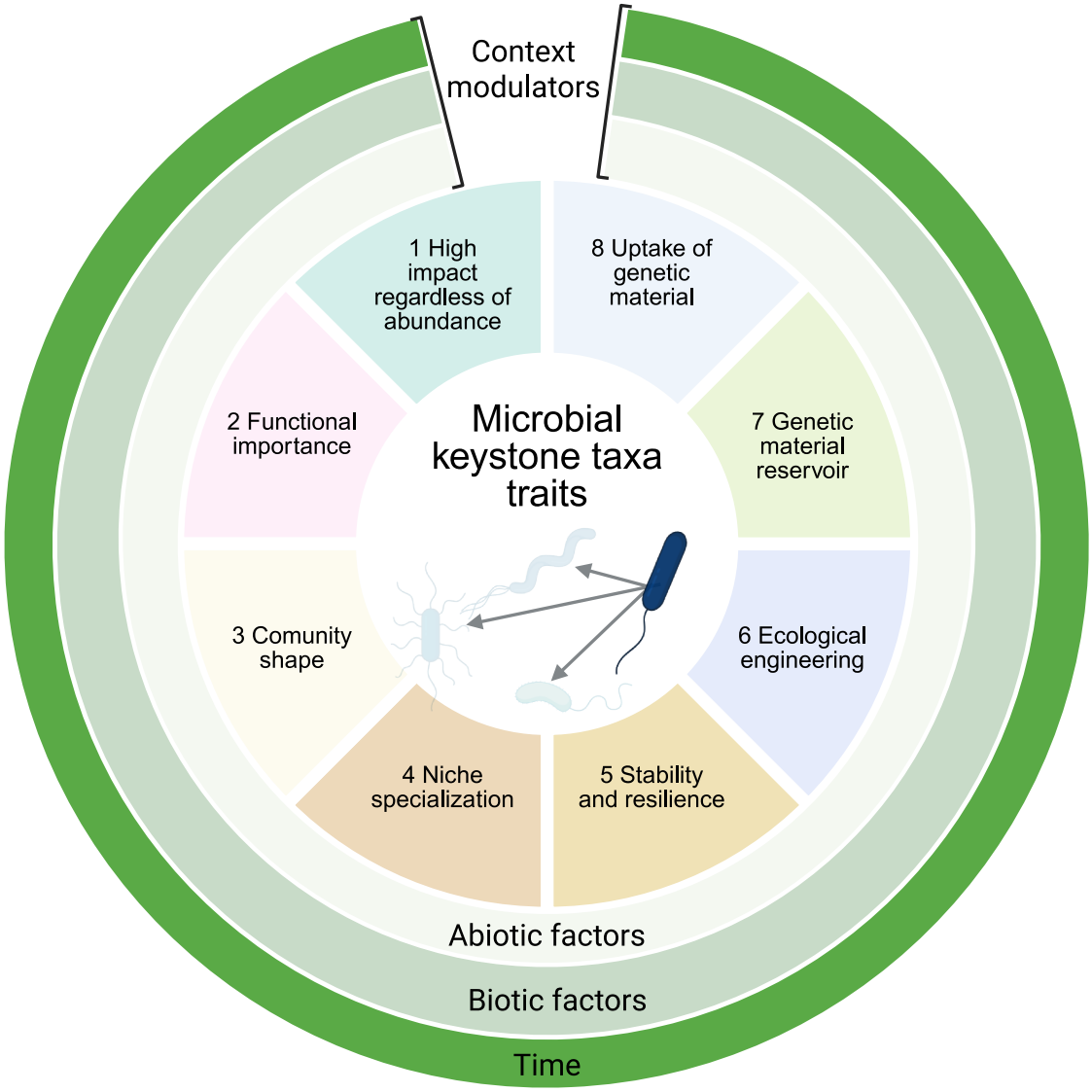

Figure 1. Trait-based framework for defining microbial keystone taxa

The concentric outer layers represent the influence of abiotic and biotic factors as well as temporal dynamics, highlighting the context-dependency of the microbial keystone status. Created in BioRender, available at https://BioRender.com/nis9mkb.

community or an ecosystem.[22] Microbial keystone taxa may exert a disproportionately large ecological impact through their activity, metabolic functions, and interactions, even when present at low relative abundance. Identifying such taxa requires high-resolution methods. Omics approaches, such as metagenomics, meta-transcriptomics, and metaproteomics, enable researchers to link microbial identity with gene content, expression profiles, and ecological traits, supporting activity-based detection of keystone functions.[23,24]

Microbial keystone taxa may contribute to essential ecosystem functions such as nutrient cycling, organic matter degradation, energy flow, pathogen suppression, and regulation of the host immune system (Figure 1, functional importance) or regulate the abundance and activity of other microorganisms within the community through various mechanisms. This regulation may involve competition, predation, symbiosis, or production of signaling molecules that modulate microbial interactions and phenotypes (Figure 1, community shape). Microbial keystone taxa may have specialized metabolic capabilities that enable them to fulfill unique or critical roles within their community, occupying specific ecological niches within their habitat and exploiting resources or environmental conditions less accessible to other organisms. Their ability to thrive in these niches may be facilitated by specialized adaptations or metabolic pathways (Figure 1, niche specialization). Consequently, their presence and activity may create microhabitats, alter nutrient availability, or modify physicochemical conditions, shaping the broader ecosystem landscape (Figure 1, ecological engineering). The

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Box 1. Definition of microbial keystone taxa

Microbial keystone taxa are specific microorganisms within a community that exert a disproportionately large impact on ecosystem structure, stability, and function relative to their abundance. They may regulate community dynamics through specialized metabolism, interactions, or ecological engineering, shaping nutrient cycling and microhabitats. Keystone status is often context-dependent, shifting in response to environmental conditions, resource availability, or community state. Unlike in macroorganisms, microbial keystone traits may vary across strains because microorganisms can acquire genes directly from their environment or from other organisms (horizontal gene transfer). Their genomes can also undergo rearrangements, gene loss, gene duplication, and other modifications that enable rapid adaptation to environmental changes (genomic plasticity) Identifying these taxa should rely on integrative omics approaches linking microbial identity with ecological function.

presence of keystone taxa enhances the stability and resilience of microbial communities. The processes behind these phenomena are still not well understood, but might be linked to larger genome sizes or highly structured genomes (Figure 1, stability and resilience). This feature might enhance their ecological roles and adaptability, not only to themselves but also to the community, as other microbiota might incorporate genetic material from the keystone taxa (Figure 1, genetic material reservoir). Vice versa, genetic material could also be transferred to keystone taxa genomes; considering that that keystone taxa are active and hold a central position in their community network, they might have an enhanced possibility to uptake genetic information from other microorganisms compared with those microbes which are less active or have lower centrality in their community network, resulting in higher adaptability under changing environmental conditions (for some examples see Zhang et al.[25] and Li et al.[26] and Figure 1, uptake of genetic material).

These eight traits (summarized in Figure 1 and Box 1) offer a practical and flexible framework to guide the identification and prioritization of microbial keystone taxa across ecosystems, linking ecological relevance to functional potential. However, these criteria are not prescriptive- some microbes that may not be classified as keystone taxa still exhibit one or more of these defining characteristics, whereas a keystone taxon does not necessarily need to fulfill all criteria mentioned. We illustrate our framework with the two showcases summarized in Box 2, describing the keystone taxa *Faecalibacterium prausnitzii* (human gut) and *Solirubrobacterales ginsenosidimutans* (soil).

ARE MICROBIAL KEYSTONE TAXA CONTEXT-DEPENDENT?

Like for many other concepts derived from the ecology of macroorganisms, applying the keystone taxa concept to the microbial world involves several potential constraints that must be considered. The limitations range from difficulties in directly observing microbial interactions to their high spatial and temporal fluctuations, as well as issues related to the exchange of genetic information via horizontal gene transfer, among others.[37] Thus, it needs to be considered whether the status of a microbial keystone taxa is permanent—inherent to the taxon itself and its environment—or it might change according to time/space variation such as nutrient availability, pH, temperature, oxygen gradients, etc.[38] In other words, does a particular ecosystem always depend on the same keystone taxa? Or might the keystone status circulate across taxa?

The current microbial keystone taxa definition does not explicitly consider the role of microbial taxa in community recovery following environmental disturbances or stress. In microbial ecosystems, some taxa may not prevent functional decline during disturbance but are essential for restoring community composition or functioning afterwards. It is important to distinguish between ecosystem stability—the ability of a community to resist change—and ecosystem recovery, which reflects the capacity to rebound and restore function after disturbance. For example, in the context of post-antibiotic recovery in the human gut microbiome, the microbial taxa facilitating the initial recovery are not necessarily those performing specific ecological functions under stable conditions. Chng et al.[39] showed that early recovery is often driven by generalist bacteria capable of degrading complex carbohydrates into simpler compounds, which in turn support the regrowth of more functionally specialized species. This suggests that taxa essential for ecosystem resilience and recovery may differ from those considered keystone under equilibrium conditions.

In addition, the dynamic activity patterns of microbes—from active to dormant states[40]—suggest that keystone status may not be a fixed trait but rather one that can shift among taxa depending on environmental conditions and ecological context. However, a certain degree of resilience to natural fluctuations is likely necessary to maintain both microbiome stability and broader ecosystem functions.

Finally, the genomes of bacteria and archaea are far more dynamic in their content and structure compared with plants and animals, often exhibiting higher rates of genetic variation, raising important questions about the long-term genomic stability of specific strains within their native environments. Indeed, laboratory experiments have shown transfers of plasmids from recipients to donors within minutes and a very fast rearrangement of genomes to cope with the applied stress.[41,42] In complex ecosystems such as the human gut, empirical studies have also revealed widespread horizontal gene transfer, suggesting that genomic exchange remains ecologically significant even outside of controlled conditions.[43] As the process of horizontal gene transfer and genome rearrangement is energy-consuming, transfer rates might be significantly lower in nature, but the dynamics of genomes of strains might still be high. Hence, when referring to microorganisms, the term keystone taxa[22] is preferred instead of keystone species, and the suitable taxonomic resolution depends on whether a function is essential for the life of a bacterial species and part of the corresponding core genome, or only present in the pangenome and harbored only by some strains.[42]

Such evidence underscores the need for a more nuanced and context-dependent definition of microbial keystone taxa—one that leaves room to integrate both ecological functions and community dynamics across temporal and environmental gradients, depending on the particular research aim. Incorporating this

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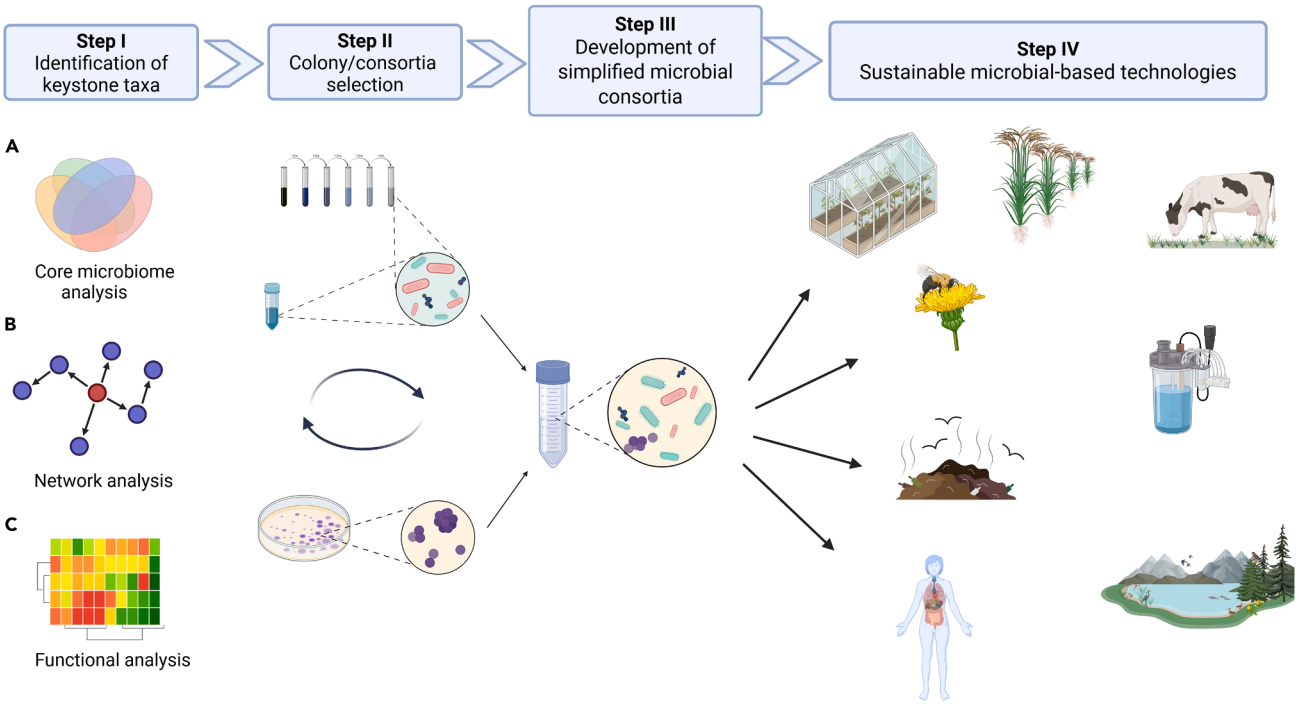

Figure 2. Identification of microbial keystone taxa and simplified microbial consortia assembly for sustainable microbiome solutions

Step I (A–C) include data-analysis techniques for identifying the environment's keystone taxa: (A) core microbiome analysis involves metabarcoding taxonomic identification to determine shared taxa among two or more microbial communities in a given host species or environment. (B) Network analysis is also usually based on metabarcoding data to determine co-occurrence associations between taxa of the microbial community, allowing the identification of hubs that strongly influence network morphology. (C) Functional analysis could include high-throughput metabolic *in vitro* profiling,[17] single-gene annotation, SIP, and, more recently, meta-transcriptomics and metabolomics, among others.[18] Step II includes a combination of isolation techniques that target the selected taxa or consortia from step I. Step III includes developing the simplified microbial consortia from strains or consortia, including development, testing, and preservation. Step IV includes the implementation of the designed simplified microbial consortia into the *in vivo* affected environment. Created in BioRender, available at https://BioRender.com/5cgn3j5.

complexity into an operational framework would not only improve our theoretical understanding but also enhance our ability to prioritize and deploy specific taxa for isolation, enrichment, and incorporation into simplified microbial consortia.

FROM DEFINITION TO IMPLEMENTATION: A TOOLBOX TO IDENTIFY MICROBIAL KEYSTONE TAXA

Still, *in silico* tools are mostly used to identify microbial keystone taxa.[44] This requires a synergistic and integrative approach, combining the power of high-throughput sequencing technologies and bioinformatics with a comprehensive understanding of microbial ecology for a particular ecosystem (Figure 2). The 4-step approach shown in Figure 2 is a flexible and integrative workflow to reflect the complexity of microbial keystone traits outlined in Box 1—linking indirect traits, such as community influence (e.g., co-occurrence, network position), with direct functional traits like metabolic activity, resilience, or ecological engineering.

Step I includes the identification of "candidate microbial keystone taxa." Network or co-occurrence analysis using metabarcoding data, based on marker genes such as the 16S rRNA gene for bacteria and archaea, the internal transcribed spacer (ITS) region of the ribosomal operon for fungi, or the 18S rRNA gene for protists, is often used for this purpose (Figure 2A). In the original study on the role of sea stars, Paine intuitively represented the shore community as a net of species connected by lines. However, formal network thinking in ecology was introduced in 2005, when Proulx and colleagues, taking advantage of the vast theoretical background from graph theory, applied the concept of co-occurrence networks to the study of ecological interactions.[45] Even though network analysis has proved to be a valuable tool for representing community structure, some aspects hinder the direct application to studying environmental microbiomes.[46–48] One relevant issue is that network analyses—based on co-occurrence or its derived associations—allow the identification of hubs with a strong influence on the network morphology. These hubs can be interpreted as "keystone taxa candidates" as their removal/loss significantly alters network morphology. However, the network represents putative ecological associations that require further assessment to gain mechanistic insights into the particular community. The co-occurrence networks may also give information about the stability of certain taxa under changing ecosystem conditions if samples from different time points are available. These microbes might be considered as part of the Core microbiome of an ecosystem (Figure 2B)[49,50] and are typically considered important for maintaining the stability, functionality, and health of an

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Box 2. Two examples of microbial keystone taxa

***FAECALIBACTERIUM PRAUSNITZII* (HUMAN GUT)**

It is one of the most abundant commensal bacteria in the healthy human colon, often representing up to 5%–15% of the total bacterial population.[27] As a prominent member of the Firmicutes phylum, *F. prausnitzii* is a major producer of the short-chain fatty acid (SCFA) butyrate. Butyrate serves as the primary energy source for colonocytes, strengthens the intestinal epithelial barrier, and modulates immune responses by promoting regulatory T cell differentiation and downregulating pro-inflammatory cytokines such as interleukin (IL)-6 and tumor necrosis factor α (TNF-α).[28,29]

Beyond direct metabolic contributions, *F. prausnitzii* engages in syntrophic interactions within the gut ecosystem. It consumes products of primary degraders of complex carbohydrates, whereas its metabolites in turn support the growth of other beneficial microbes, reinforcing a stable and resilient microbial network. Reduction in *F. prausnitzii* abundance is consistently observed in dysbiosis associated with inflammatory bowel disease (IBD), obesity, type 2 diabetes, and other chronic conditions, highlighting its central role in maintaining eubiosis.[30]

Thus, although it may not fulfill all trait-based components, *F. prausnitzii* is an example of a keystone taxon as it shows high impact on microbial community composition (Figure 1, high impact regardless of abundance), its presence sustains critical gut functions (Figure 1, functional importance)—nutrient metabolism, barrier integrity, and immune homeostasis—whereas its loss destabilizes the microbial community and predisposes the host to disease (Figure 1, stability and resilience).[31,32] Targeted strategies to preserve or restore *F. prausnitzii* populations, such as prebiotic interventions, next-generation probiotics, or microbiota transplantation, are being actively explored to harness its role in gut health.[28–30]

***SOLIRUBROBACTERALES GINSENOSIDIMUTANS* (SOIL)**

It is characterized by its metabolic versatility and ability to degrade complex organic compounds, including plant-derived saponins such as ginsenosides, which promote beneficial plant-soil feedback loops by mitigating the accumulation of inhibitory secondary metabolites that would otherwise suppress plant growth and soil fertility.[33,34] *S. ginsenosidimutans* also contributes to soil network connectivity and soil ecosystem services stability, as maintaining a steady flux of carbon substrates supports syntrophic interactions with other microbial guilds, including nitrifiers, denitrifiers, and mycorrhizal fungi.

Declines in its abundance are correlated with reduced soil fertility, increased disease susceptibility in crops, and disrupted nutrient cycling. Studies based on high-throughput sequencing and co-occurrence network analysis reveal that this taxon often occupies hub positions in microbial interaction networks,[35] suggesting that its presence stabilizes community structure against environmental perturbations.[36]

Thus, *S. ginsenosidimutans* is an example of a soil keystone taxon that combines specialized metabolism and community-wide functions. Its metabolic outputs sustain multiple trophic levels of the soil microbiome (Figure 1, high impact regardless of abundance), through the secondary metabolite transformation (Figure 1, functional importance), the niche specialization in the ginseng rhizosphere (Figure 1), and ecological engineering capacity (Figure 1). Future research on its interactions, genomic adaptations, and bioinoculant potential could support soil health and crop productivity under global change

ecosystem facing natural dynamics in chemical and physical properties, an essential feature of a keystone taxon (see above). However, in contrast to macroorganisms, where these interactions are visible and functional dependencies between organisms can be easily assessed, the microbial world network analysis is a purely statistical analysis, and links to functions are missing. Therefore, a functional annotation of the "candidate microbial keystone taxa" is needed. Functional analysis (Figure 2C) could include high-throughput metabolic *in vitro* profiling,[17] single-gene annotation, stable isotope probing (SIP), and, more recently, metagenomics, meta-transcriptomics, metabolomics, genome-scale metabolic modeling,[51] among others.[18] These approaches provide insights into microbiome biosynthetic pathways and metabolic activities, contributing to understanding the functional roles of specific microorganisms within a community or environment. "Candidate keystone taxa" inferred from metabarcoding data can then be mapped with functional data, for example, by mapping to metagenome-assembled genomes (MAGs). MAGs can be used to characterize the functional potential of the identified keystone taxa and to define specific strategies for targeted isolation of the microbiota of interest.

Step II includes the subsequent targeted isolation of the candidate keystone taxa. Developing appropriate cultivation media and employing helper strains to support microbial growth are key strategies that can be guided by the functional mapping framework described earlier. However, the feasibility of isolating keystone taxa as individual strains remains a matter of debate, as their functional traits may be context-dependent and not fully expressed outside of their native community environment. Indeed, recent work by Northen et al.,[52] Mehlferber et al.,[53] and Delgado-Baquerizo et al.[54] emphasizes that isolation alone may fail to recapitulate the full phenotypic expression or ecological function of keystone taxa, suggesting the use of co-cultivation systems, synthetic consortia, microcosms, and *in situ* assays to preserve ecological interactions during controlled testing. We therefore consider both single-strain isolation and enrichment of consortia as complementary strategies: while isolating a single strain allows precise functional testing, cultivating simplified or native consortia may better preserve key interactions and unlock traits that only emerge in a community context. This dual approach may also extend the range of microbes accessible to functional characterization and practical application.

After successful isolation, strains or consortia should undergo functional validation using *in vitro* and *in vivo* models in accordance with Koch's postulates (1893). Although originally intended for pathogenic microorganisms, the principle remains relevant for characterizing other microbes as well. However, we argue that a "community-level Koch's postulate" is now necessary, accounting for microbial interdependencies and emergent functions within microbial consortia.

Furthermore, the assembly of simplified microbial consortia must consider not only the physiological and functional traits of keystone taxa but also the environmental conditions under which those traits are expressed. Strategies such as targeted metabolomics, the integration of ecological theory (e.g., niche complementarity and facilitation), and enhancing taxonomic and functional diversity within simplified microbial consortia

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can improve community stability and performance.[52,54] These approaches collectively offer a more ecologically realistic and experimentally feasible pathway toward identifying and deploying keystone taxa within microbiome engineering efforts.

Step III includes the simplified microbial consortia development, which combines selected strains or consortia at specific proportions, generally *in vitro*, under controlled conditions. The design of the simplified microbial consortia should consider not only the strains' requirements, i.e., nutrient availability, physicochemical conditions, etc., but also the interactions between strains, which determine the functionality of a simplified microbial consortia to a large extent. Although synergies between strains may be essential to obtain the proposed activity pattern of a simplified microbial consortia, and some genes might be expressed only if a certain combination of microbes is present, at the same time, negative interactions need to be considered as microbes may also interfere with each other, in a way that growth and/or activity is reduced or even completely blocked, e.g., by the formation of substances with antimicrobial properties by single members of the simplified microbial consortia. This third step might imply a loop of sub-steps of testing and tuning the culture conditions, namely the design-build-test-learn cycle (DBTL).[55] This step should include testing and preservation sub-steps.

Step IV, the final step, corresponds to the *in vivo* implementation of the established simplified microbial consortia into applications. Any simplified microbial consortia application must be considered as an invasion of microbes into an ecosystem. Thus, invasion theories derived from the ecology of macroorganisms may help determine the inoculation's success in advance. In plant seeds, for example, due to the priority effect, simplified microbial consortia strains have shown a low environmental invasion capacity; however, they can modify the seed microbiota composition, community size, and overall seedling.[56] In general, implementations of simplified microbial consortia are more likely to be successful when applied after a significant disturbance or when the environment is naturally low in microbial diversity.[57] This principle has already been implemented into clinical practice when using fecal transplants, although these fecal transplants contain a native and, sometimes, unknown microbiota rather than systematically defined and targeted simplified microbial consortia.[58] For complex ecosystems like soil, using carrier materials to protect the inoculated microbes from grazing and allow for a stepwise adaptation to the new conditions can increase the success rate for colonization of an ecosystem after introducing single microbial strains or simplified microbial consortia. Carriers used successfully for soils can be of organic (e.g., compost, biogas slurry, crushed corn cobs, biochar, peat, etc.) or inorganic origin (e.g., zeolite, perlite, lignite, talc, etc.).[59,60] The final phenotype of the simplified microbial consortia *in vivo* also depends on environmental conditions and can only be reliably predicted for housekeeping genes or constitutively expressed genes. By contrast, for most of the highly regulated genes linked to the synthesis of beneficial substances or the degradation of harmful substances—phenotypes must be assessed depending on the chemical and physical conditions of the ecosystem—thereby hindering accurate prediction of the resulting phenotype.

OUTLOOK

Recent advances in microbiome research have highlighted the promise of *in vivo* approaches that exploit microevolution and ecological filtering to identify microbial taxa with desirable functional properties. Selective pressures in natural or semi-natural environments can enrich for strains that contribute disproportionately to specific functions, providing a pragmatic route for the design of simplified microbial consortia with targeted applications.[54,55] Future research should develop *in vivo* ecological selection and microevolutionary enrichment as systematic tools to identify and validate microbial keystone taxa. Such taxa often act as central nodes within interaction networks, shaping both the stability and emergent functions of communities. By leveraging ecological selection *in situ*, we may be able to reveal those taxa most relevant for sustaining functionality under context-specific conditions that are difficult to replicate *in vitro*.

Integrating this ecological and evolutionary perspective with a trait-based framework offers several advantages. First, it allows for a more nuanced understanding of the ecological roles of candidate strains, moving beyond the presence or absence of functions toward quantifying their contributions to community resilience, robustness, and productivity. Second, coupling *in vivo* enrichment with controlled laboratory validation could establish a translational pipeline, whereby naturally selected candidates are systematically incorporated into synthetic consortia and evaluated for their biotechnological potential. This iterative cycle of ecological filtering, trait-based assessment, and experimental validation could significantly accelerate progress in microbiome engineering.

This approach also provides a pathway for aligning ecological theory with applied microbiology. Concepts such as niche complementarity, functional redundancy, and keystone taxa dynamics—traditionally studied in complex natural ecosystems—can be operationalized within simplified consortia designed for practical applications. By allowing natural environments to serve as filters, we may overcome current limitations in predicting which taxa are most critical for community function, thus reducing reliance on trial-and-error assembly strategies.

CONCLUSIONS

In summary, identifying and understanding microbial keystone taxa is crucial for developing simplified microbial consortia and unlocking their potential in microbiome engineering applications across diverse fields, including agriculture, bioremediation, and human health. We propose a trait-based 8-component framework for identifying microbial keystone taxa, linking each characteristic to its ecological significance and highlighting its unique roles within microbial ecosystems. To prioritize microbes for isolation and simplify microbial consortia development, we suggest a 4-step operational approach that integrates cultivation-based techniques with methodologies that bring information about the structure and functionality of the microbial community. Successfully implementing simplified microbial consortia designed to evaluate the role of a candidate keystone taxa could confirm its status as a keystone taxon.

CONSORTIA

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DECLARATION OF INTERESTS

The authors declare no competing interests.

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